DESIGN, CONSTRUCTION AND COMMISSIONING OF BIOREACTOR EXPERIMENTAL RIG TO MEASURE K_La OF OXYGEN IN NEWTONIAN FLUID

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ABSTRACT

In this research, a bioreactor experimental rig was designed, constructed and finally commissioned in order to measure the k_La and ka_p of oxygen in Newtonian fluid. Distilled water was used as a sample of Newtonian fluid. The design of the rig was based on the standard geometry of a stirred tank reactor. Standard geometrical ratios of a stirred tank reactor with two Rushton disc turbine (RDT) impellers was used to design a system that can provide good mixing for a gas-liquid system. Two sets of RDT impellers were used to disperse the gas sparged by a ring sparger into the liquid content in the tank. The construction of this rig was done with the help of the Assistant Training Vocational Officer in the engineering workshop. The tank was built using transparent Polyvinyl chloride (PVC) while the two impellers and four equally spaced baffles were constructed using stainless steel. The effect of two variables, namely impeller speed and air flow rate on the volumetric transfer coefficient of oxygen from gas to the bulk liquid, k_La and on the mass transfer coefficient of oxygen from bulk liquid to the oxygen electrode, ka_p were studied. From the result, the increase in impeller speed and air flow rate will both increase the value of k_La and ka_p. The values of k_La and ka_p were predicted by fitting the mass transfer equation of oxygen to the experimental data by using sum of squared error minimization of the difference between the actual and predicted data using MATLAB programming.

ABSTRAK

Dalam kajian ini, sebuah bioreaktor rig eksperimen direka, dibina dan akhirnya ditugaskan untuk mengukur k_La dan ka_p oksigen dalam bendalir Newton. Air suling digunakan sebagai sampel bendalir Newton. Rekabentuk rig adalah berdasarkan geometri piawai sebuah tangki reactor berpengaduk. Nisbah geometri piawai untuk sebuah tangki reaktor berpengaduk dengan dua pengaduk turbin cakera Rushton (RDT) telah digunakan untuk membina sebuah sistem yang dapat menyediakan percampuran yang baik untuk satu sistem gas-cecair. Dua set pengaduk RDT digunakan untuk menyebarkan gas yang disemburkan oleh penyembur cincin ke dalam kandungan cecair dalam tangki. Pembinaan rig ini dilakukan dengan bantuan daripada Pegawai Pembantu Latihan Vokasional di bengkel kejuruteraan. Tangki dibina menggunakan PVC telus sedangkan dua pengaduk dan empat bafel sama ruang dibina menggunakan keluli tahan karat. Pengaruh dua pembolehubah iaitu kelajuan pengaduk dan kelajuan aliran udara terhadap pekali pemindahan isipadu oksigen dari gas ke cecair pukal, k_La dan pekali pemindahan jisim dari cecair pukal ke elektrod oksigen, ka_p telah dikaji. Hasilnya, dari kajian ini, didapati bahawa peningkatan kelajuan pengaduk dan kelajuan aliran udara kedua-duanya meningkatkan nilai k_La dan ka_p. Penganggaran nilai k_La dan ka_p dilakukan dengan cara menyesuaikan persamaan pemindahan jisim oksigen kepada data yang diperolehi melalui eksperimen dengan meminimumkan jumlah ralat ganda dua perbezaan diantara data-data sebenar dengan data-data yang telah dianggarkan menggunakan pengaturcaraan MATLAB.

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LIST OF ABBREVIATIONS AND SYMBOLS

ρ	-	Liquid density (kg/m^3)
μ	-	Liquid viscosity (Pa.s)
σ	-	Standard error
a	-	Specific surface of interface
B	-	Baffles width
C	-	Bottom clearance
C*	-	Saturated dissolved oxygen concentration (8.13 ppm)
C_L	-	Dissolved oxygen concentration in bulk liquid
C_p	-	Dissolved oxygen concentration at the probe
D_i	-	Impeller diameter
D_t	-	Tank diameter
DO	-	Dissolved oxygen
DOC	-	Dissolved oxygen concentration
DOT	-	Dissolved oxygen tension
F(t)	-	Function to be minimized
F_r	-	Froude number
g	-	Gravitational acceleration (9.81 m/s^2)
H_L	-	Liquid height
H_i	-	Height between impellers
H_t	-	Tank height
ka_p	-	Electrode mass transfer coefficient
k_La	-	Volumetric mass transfer coefficient
L	-	Length of impellers blade
N	-	Impeller speed
N_{CD}	-	Impeller speed for complete dispersion

N_F	-	Impeller speed for flooding
N_{po}	-	Ungassed power number
N_Q	-	Discharge coefficient
N_R	-	Impeller speed for recirculation
N_{Re}	-	Reynold's number
P_o	-	Ungassed power consumption
P_g	-	Gassed power consumption
PVC	-	Polyvinyl chloride
Q_g	-	Gas flow rate
R^2	-	Coefficient of determination of linear regression
RDT	-	Rushton disc turbine
t	-	time
W	-	Width of impellers blade
x1	-	Initial guess of ka_p
x2	-	Initial guess of k_La
YR(t)	-	Theoretical dissolved oxygen concentration at time t
YS(t)	-	Experimental dissolved oxygen concentration at time t

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Oxygen is a key factor for biological activity in a bioreactor where aerobic microorganisms presents. For example, in aerobic fermentation process, the supply of molecular oxygen to the surface of each microbial cell is the primary importance. The dissolved oxygen in the liquid medium is utilized for the microbial growth, product formation, and provision of energy through the respiration of the microorganisms. The study of transport process of oxygen in a gas-liquid system is important to determine how the dissolved oxygen is transferred to the liquid phase of the system. In such mixing phenomena, the study of oxygen transfer rate to the liquid medium is crucial for the optimization of the process.

Since oxygen is sparingly soluble in water, it may be the growth-limiting substrate in these fermentations. For bacteria, the critical oxygen concentration is about 5% to 10% of the saturated dissolved oxygen concentration, (DOC). Above this critical concentration, the oxygen concentration no longer limits growth (Shuler and Kargi, 2002). For optimum growth, it is therefore important to maintain the DOC above this critical level by sparging (bubbling gas through) the fermenter with air or pure oxygen. Of course, to be effective, the mass transfer rate from the gas bubbles to the liquid broth must equal or exceed the rate at which growing cells take up the oxygen.

Oxygen transfer from gas bubbles to the cells is usually limited by oxygen transfer through the liquid film surrounding the gas bubbles. The rate of oxygen transfer from the gas to the liquid phase is given by the equation

$$OTR = k_La(C^* - C_L) \qquad (1)$$

where k_La is volumetric oxygen transfer coefficient, C^* is saturated dissolved oxygen concentration and C_L is actual dissolved oxygen concentration in the bulk liquid. Also, OTR is the term for oxygen transfer rate.

The kLa include two factors: (i) the mass transfer coefficient k_L which is mainly a function of the physical properties of the system and the temperature (O'Connor, 1955); and (ii) the specific surface of the interface ``a". Operation with smaller bubbles size is preferred as it can provide greater specific surface area of interface per unit volume. On the other hand, the concentration difference (C^* - C_L) is not significantly influenced by the physical system and it depends both on the oxygen concentration in the air inlet and the temperature of the operation (Levonen and Llaguno, 1984). C^* decrease as the temperature of operation increases. Taking all these issues into consideration, it seems clear that it is necessary to know the values of k_La because it has been regarded as the most appropriate parameter to characterize oxygen transfer in a particular system (I. de Ory *et al*, 1999).

In University Malaysia Pahang, the students in Faculty of Chemical and Natural Resources Engineering study the effect of several variables on k_La value by utilizing a 10L fermenter that is available in the bioprocessing laboratory. The improper commission of the experiment by the undergraduate students has lead to the damage of the fermenter. This phenomenon has caused several difficulties to the postgraduate students and the fourth year students who want to use the fermenter for their research and fermentation process. In addition, sometimes the utilization of this fermenter during lab session is restricted because it has been booked or in used by the postgraduate student or the fourth year students who want to run their research. In order to encounter this problem, a new bioreactor experimental rig must be designed and constructed to be utilized by the undergraduate students in order to measure the k_La value during their lab session.

The design and construction of this experimental rig has two main significances. The first one is for students comfort in study. Currently, the undergraduate students have to share the 10L fermenter available in the lab with the postgraduate students. This new rig will help the undergraduate students to study the k_La during their lab session without any intersection with the postgraduate research. In addition, the research done by the postgraduates and the fourth year students in their research project will not be interrupted by the lab session. Furthermore, the invention of this rig will enhance the future study on other operating variables that can affect the k_La value such as type of impeller, non-Newtonian fluids, the presence of alcohol, inorganic electrolytes and antifoaming agent. In other words, the design and construction of this bioreactor experimental rig will contribute to the research field and student comforts in study.

1.2 Problem Statement

Since the undergraduate students in Faculty of Chemical and Natural Resources Engineering are using the fermenter in the bioprocess laboratory to run their experiment to study the effect of several parameters on k_La value, they have to share the equipment with the postgraduate students who use the fermenter for fermentation process. Unfortunately, sometimes they cannot run their experiment because the device is in use or have been booked by the postgraduate students for their research.

Recently, faculty spent a lot of money for maintenance of the fermenter. This is due to the improper commissioning of the fermenter by undergraduate students which caused several damages to the device. This is such a waste of money. Besides, the fermenter will be unavailable to be used by the postgraduate students because of maintenance. This incident will disturb the progress of the postgraduate research and result in the alteration of the lab session schedule.

In order to design a new rig for experimental studies, there are several problems need to be considered about the oxygen transport phenomena in the process. Firstly, the mass transfer of oxygen in the process will encounter several barriers. These barriers need to be reduced by appropriate geometrical design of the rig in order to provide excellent gas-liquid mixing.

For any fermentation process to be conducted, it is necessary to determine the volumetric mass transfer coefficient (k_La) of oxygen value in order to study the oxygen transfer characterization in the liquid medium for the optimization of the process. The k_La value for oxygen is influenced by several operating variables such as intensity of agitation and the aeration rate. Thus, it is necessary to determine the effect of these operating variables on the k_La value prior to any fermentation process.

1.3 Objectives

The purpose of doing this study is to achieve following objectives:

1) To design and construct a bioreactor experimental rig that can enhance gas-liquid mixing for transport process of oxygen in Newtonian fluids in order to be utilized by undergraduate students in their experiments for the determination of k_La value during lab session.

2) To study the effect of air flow rate and impeller speed on k_La and ka_p value for oxygen in Newtonian fluids.

1.4 Scopes of Study

In order to achieve the objectives of this project, the following studies will be carried out:

1) Theory of oxygen transfer process.
2) Standard geometrical design for a stirred tank reactor.
    - Standard geometrical design of stirred tank
    - Standard geometrical impeller design
    - Liquid flow patterns
    - Turbulence
3) Power consumption in stirred tank
    - Single Newtonian liquid
    - Reduced power consumption in aerated tank
    - Gas flow patterns
4) Operation variables affecting k_La and ka_p value.
    - Impeller speed
    - Air flow rate
5) Methods to determine volumetric mass transfer (k_La) of oxygen.
    - Static gassing out and other methods (Experimental)
    - Sum squared error minimization analysis (MATLAB)

CHAPTER 2

LITERATURE REVIEW

2.1 Theory of Oxygen Transfer Process.

Most industrial microbial processes are aerobic, and are mostly carried out in aqueous medium containing salts and organic substances; usually these broths are viscous, showing a non-Newtonian behavior. In these processes, oxygen is an important nutrient that is used by microorganisms for growth, maintenance and metabolite production, and scarcity of oxygen affects the process performance (Garcia-Ochoa *et al.*, 2000a; Calik *et al.*, 2004; Liu *et al.*, 2006a). Therefore, it is important to ensure an adequate delivery of oxygen from a gas stream to the culture broth.

In many chemical and biochemical reaction, aeration is needed to supply oxygen that will be consumed in the particular reaction pathways. The oxygen can be supply to the system by means of air compressor or sparging pure molecular oxygen. In aerobic fermentation process, the supply of molecular oxygen to the surface of each microbial cell is very crucial in order for the cell to absorb the oxygen molecule to be consumed. Before reaching the surface of the microbial cell, a transport process will occur along the pathway from the air bubbles through the liquid medium. For the high cell density culture, problem arises where the growth of the microbial cells is limited by oxygen availability in the liquid phase. The mass transfer process of molecular oxygen in this system will encounter many types of barriers.

In order to make the oxygen is available for the microorganism, it must be bring into contact with the liquid and then transferring the dissolved gas from the gas-liquid interface to the microorganisms. Rapid oxygen supply needs large surface area of contact between the gas and liquid to facilitate the dissolution and turbulence and finally mix the dissolved gas into the bulk of the fermentation. Figure 2.1 describes the transport process of oxygen graphically.

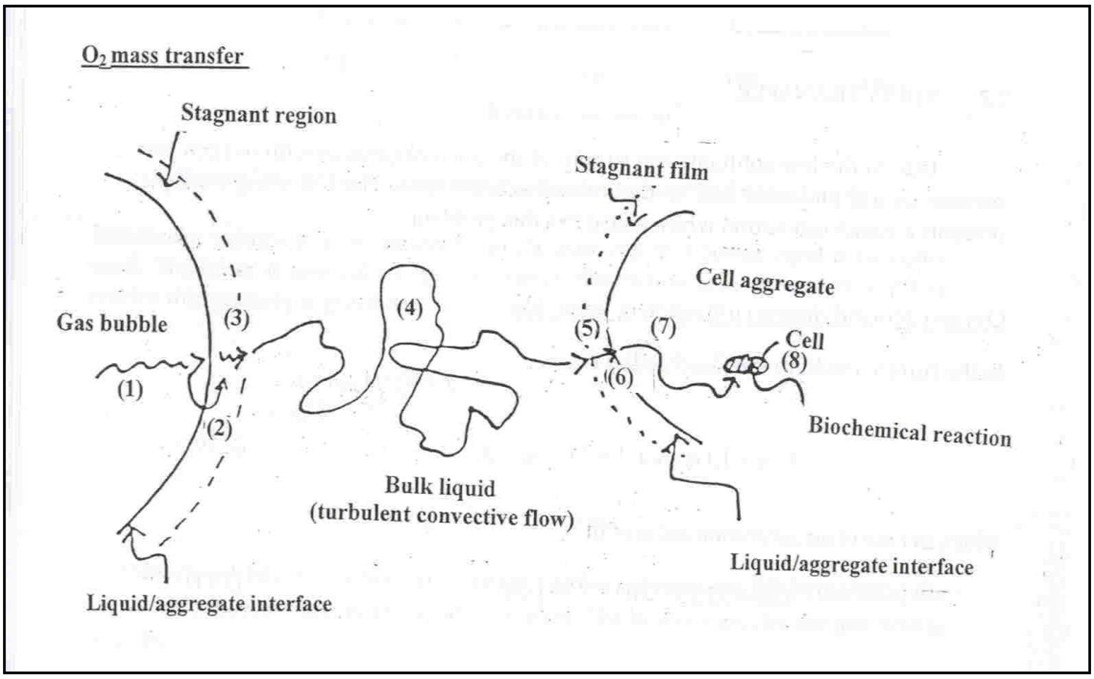


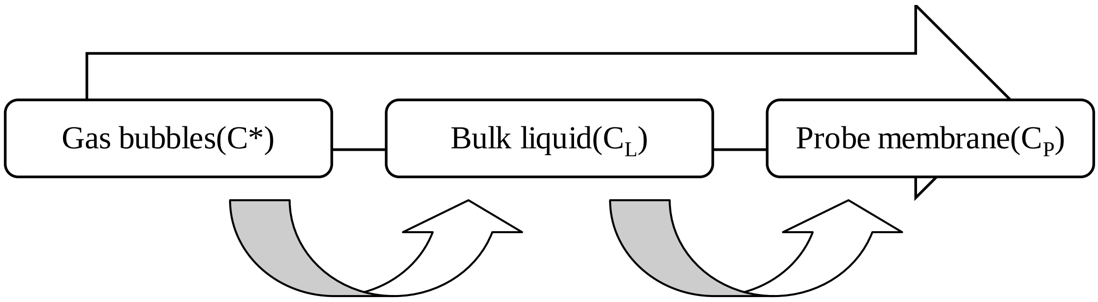
Figure 2.1 : Graphical view of oxygen transport process

From the figure shown above, we notice that the mass transfer process will encounter several barriers:

1) Gas film resistance between bulk gas and gas-liquid interface
2) Interfacial resistance at the gas-liquid interface
3) Liquid film resistance between interface and bulk liquid phase
4) Liquid phase resistance for oxygen transfer to the liquid film surrounding a single cell
5) Liquid film resistance around the cells
6) Intracellular or intrapellet resistance (for microbial flocs or pellets)
7) Resistance due to consumption of oxygen inside the microbial cells

The sum of all resistance is equal to the overall resistance to transfer the oxygen into the cell. The magnitudes of individual resistance depend on the bubble and liquid phase hydrodynamics, composition and rheological properties, densities, cell activities and gas- liquid interfacial phenomena. The sum of gas film, liquid film and interfacial resistances to oxygen transfer is reciprocal of overall oxygen transfer coefficient.

In this study, the mechanism of oxygen transfer is the same but the research is conducted using distilled water as a Newtonian fluid. Thus, the oxygen transport process can be illustrated by:



Theoretically, oxygen transfer process can be described as follow (Winkler, 1981) : The air is sparged into the distilled water content in the stirred tank. Oxygen from the gas bubbles will be transferred into the bulk liquid.

$$dC_L/dt = k_La(C^*-C_L) \tag{2}$$

Then, the dissolved oxygen from the bulk liquid will be transferred to the oxygen electrode's membrane.

$$dC_p/dt = ka_p(C_L-C_p) \tag{3}$$

After integration and rearrangement of equation, the theoretical dissolved oxygen concentration can be correlated by (Asis, 1990)

$$YR(t) = C^*-C^*[(ka_p.\exp(-k_La.t))/(ka_p-k_La) – (k_La.\exp(-ka_p.t))/(k_La-ka_p)] \tag{4}$$

where YR(t) = theoretical oxygen concentration at time t

C* = Saturated oxygen concentration in bulk liquid (8.13 ppm)

kap = electrode mass transfer coefficient of oxygen

kLa = volumetric mass transfer coefficient of oxygen

t = time

2.2 Standard Geometrical Design

2.2.1 Standard Geometrical Design of a Stirred Tank

In chemical processing, there is no such thing as a standard geometry stirred tank. However, most design information, from experimental studies and plant scale measurements, exist for the range of geometries given in table 2.2.1. Fig 2.2.1 shows the geometry of stirred tanks and defines the important dimensions. Many research has been carried out in flat bottomed tanks, despite the fact that the majority of industrial vessels have dished ends (ellipsoidal or torispherical) for the ease of fabrication and cleaning, and operation at elevated pressure.

Table 2.1 : Geometrical ratios of a stirred tank

	Geometric Ratio	Typical Range	Standard Geometry
liquid height	H / T	1-3	1
impeller dia	D / T	1/4 - 2/3	1/3
bottom clearance	C / T	1/4 - 1/2	1/3
bottom clearance	C / D	~1	1
baffle width	B / T	1/12 – 1/10	1/10

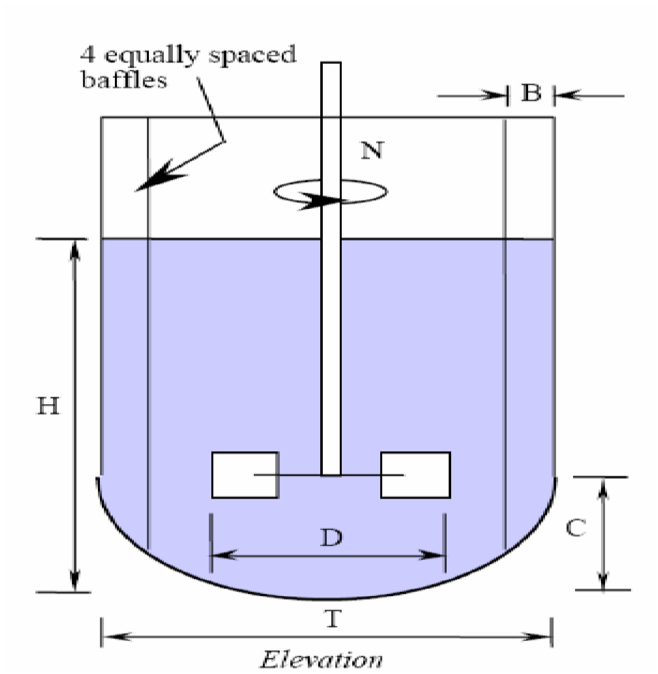


Figure 2.2 : Geometry of a stirred tank

The tank may be baffled or unbaffled. More effective mixing is obtained by placing baffles on the tank wall which generate large axial and radial velocities rather than a purely swirling flow. Full baffling may be achieved using four vertical baffles mounted radially, 90˚ apart; the baffles should extend to at least the free surface but often have a small clearance from the base of the vessel. For fluid mixing with dispersed solid particles, the baffles may be supported off the wall, leaving gap of ~ T/14. This is designed to prevent build-up of particles in the crevice between the baffles and the wall and to facilitate cleaning.

2.2.2 Standard Geometrical Impeller Design.

In low viscosity liquids, smaller diameter impellers (small D/T ratios) are able to generate flow in all parts of the tank at moderate power input. The common impeller types and its standard geometries are shown in figure 2.2 and can be classified according to the type of discharge flow produced. Figure 2.3 shows regular impellers dimension.

Table 2.2 : Impeller flow types and examples

Impeller flow type	Examples
Radial	Flat paddle, disc turbine
Axial	Marine propeller
Axial and radial mixed flow	Pitched-blade turbine, hydrofoil

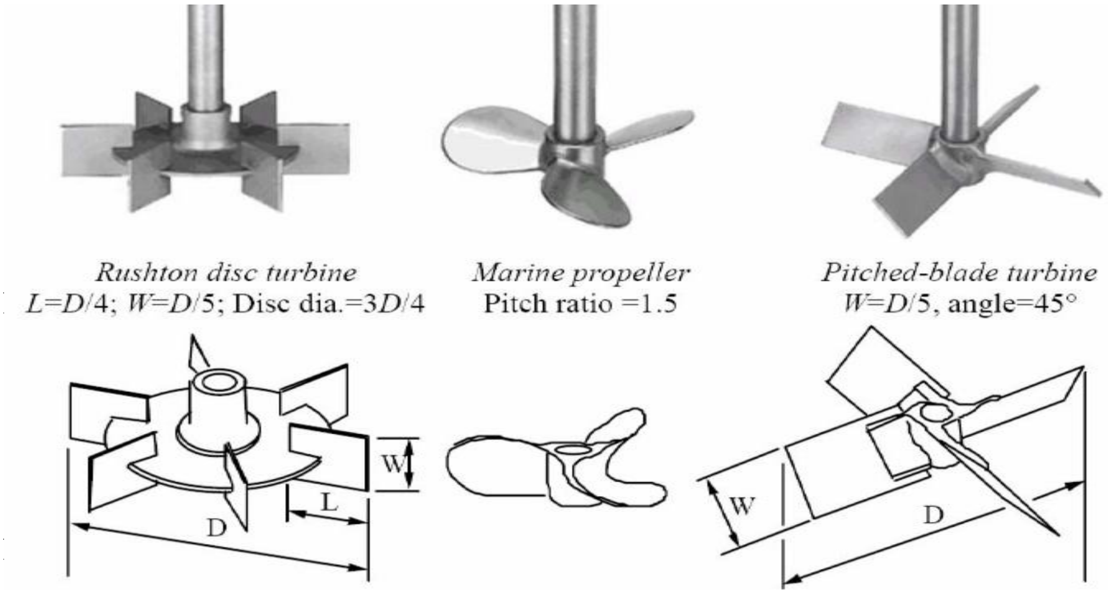


Figure 2.3 : Regular impeller dimension

With aspect ratio (H/T) greater than about 1.5, it is usual to have multiple impellers on the same shaft (each distance of ~ 1-2D apart) to give effective agitation throughout the tank volume. These impellers also have standard geometry design, e.g. a typical width to diameter ratio, W/D of 1/5 for Rushton disc turbine and mixed-flow pitched-blade turbines. A large number of literature measurements have been made on the standard Rushton disc turbine (6 bladed); formerly this design was regarded as one of the best multi-purpose agitators. However, recent research has shown that hydrofoil or pitched-blade impellers have certain advantages for specific low viscosity operation. Figure 2.4 shows some mixer selection chart for fluid processing.

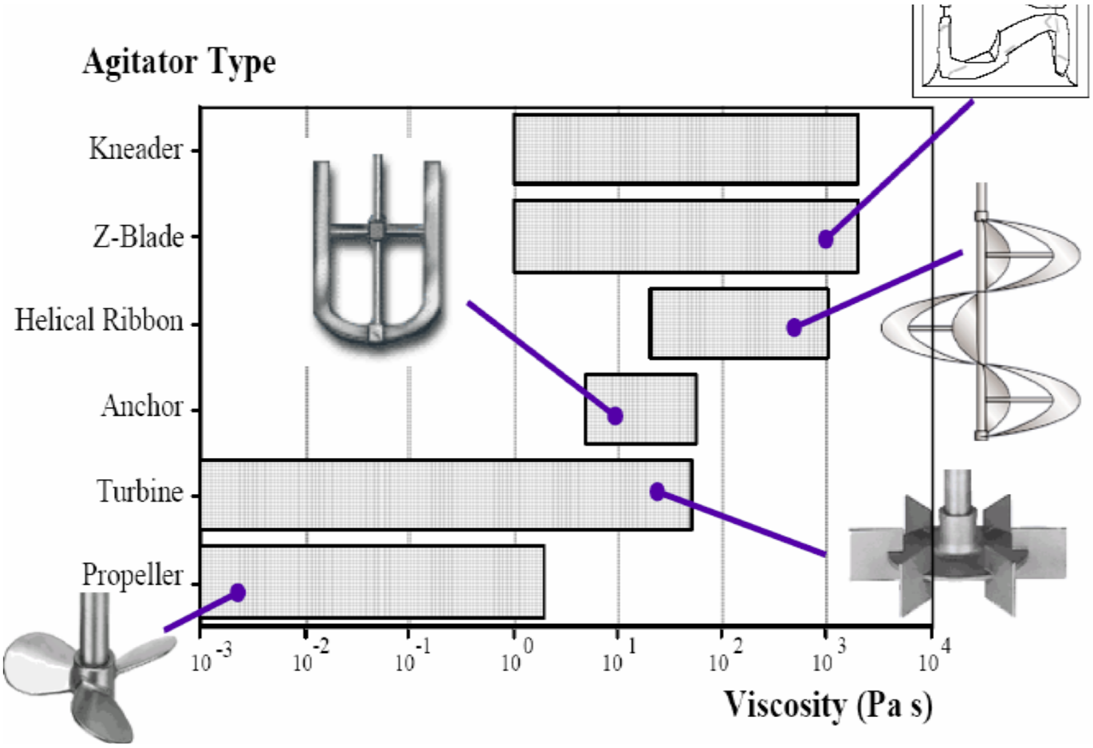


Figure 2.4 : Mixer selection chart for fluid processing

2.3 Liquid Flow Patterns

All small diameter impellers, rotating at high speed in low viscosity liquids, in unbaffled tanks produce a predominantly tangential swirling flow, with weaker, secondary vertical circulations. (Nagata, 1975) presents velocity profiles for a variety of impellers in unbaffled tanks and describes a theoretical model for the flow, consisting of a central solid body (forced vortex) region with an outer free vortex. In the central, solid body rotation there is no relative movement of fluid elements and hence no mixing. In the outer region, mixing is only achieved in the tangential direction. At higher impeller speeds, the surface vortex extends to the impeller blades and air is entrained. Consequently, unbaffled tanks are not efficient for blending operations. The use of an eccentric impeller improves blending efficiency by preventing the formation of the forced vortex and also introduces an additional problem that air is entrained from the free surface at low impeller speeds.

Baffling redirects the tangential and radial flow in the impeller discharge stream, generating strong vertical circulations. In baffled tanks, two distinct types of flow may be identified: 1) radial flow, as produced by the Rushton disc turbine; and, 2) axial flow, as produced by the marine impeller. Pitched-blade turbines generate a mixed flow with both axial and radial components in the discharge stream. These common flow patterns are illustrated in figure 2.5. In addition, there is evidence [Yianneskis *et al*., 1987] to show that these flows exhibit pseudo-periodicity due to shedding of trailing vortices from the impeller blades which means there is an element of unsteadiness in the flows.

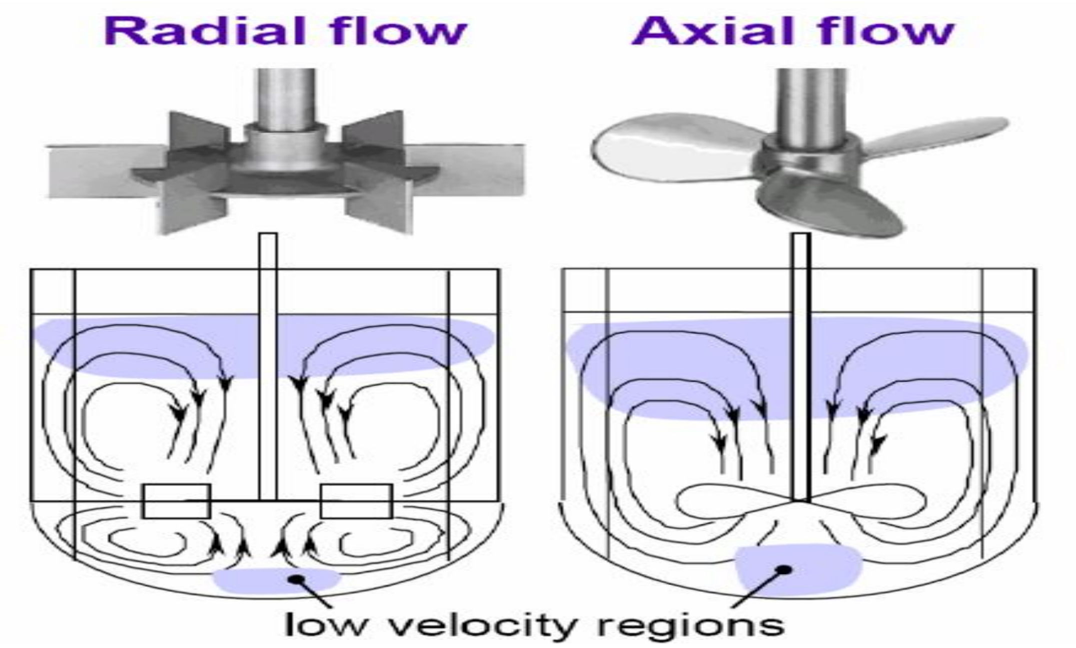


Figure 2.5 : Common flow patterns produced by impellers

More modern impeller designs like hydrofoil impeller give various combinations of axial and radial flow, depending on blade shape and pitch. In blending process, the impeller should produce a strong circulating flow but consume only small amount of power. In contrast, in gas-liquid or liquid-liquid dispersions, high rates of energy dissipation are required to break up droplets or bubbles. Recent interest has focused on the distribution of energy dissipation within the tank because the energy input through the shaft is not dissipated uniformly throughout the tank. For example, the radial disc turbine has a high power input and also has high rates of